

PHYTOCHEMICAL EVALUATION OF SEED AND FRUIT PULP EXTRACTS OF *PASSIFLORA FOETIDA* L.

P. S. Chinnasamy, S. Parimala and M. Kandhasamy*

PG and Research Department of Botany Government Arts College (Autonomous), Karur -
639005.

Article Received on
14 Feb. 2018,

Revised on 07 March 2018,
Accepted on 28 March 2018,

DOI: 10.20959/wjpr20187-11770

*Corresponding Author

M. Kandhasamy

PG and Research

Department of Botany

Government Arts College

(Autonomous), Karur -

639005.

ABSTRACT

The methanolic extracts of seed and fruit pulp were examined for possible sources of phytochemical constituents. The preliminary phytochemical screening was done with standard qualitative procedures which revealed that the presence of several secondary metabolites. GC-MS study was made to identify the potential bioactive compounds present in the seed and pulp extracts. GC-MS analysis showed the presence of 12 and 15 bioactive compounds in seed and fruit pulp extracts respectively. The most prevailing major compounds present in seed extract were, 11-Octadecenoic acid, methyl ester (31.82 %), 9, 12-Octadecadienoic acid, methyl ester (27.08 %), Hexadecanoic acid, methyl ester (21.94 %), Hexadecanoic acid, 15methyl-, methyl

ester (13.65 %) and Phthalic acid, isobutyl octyl ester (2.85 %) and the methanolic extract of fruit pulp showed the presence of Hexadecanoic acid, 15-methyl-, methyl ester (26.39%), 5Hydroxymethylfurfural (25.18%), 2',3'-Dideoxyribonolactone (11.89%), Levoglucosenone (11.21%), Furfura (6.76%). These results indicated that the methanolic extract of seed and fruit pulp of *P. foetida* and hence possess potent antibacterial, antioxidant, anti-inflammatory, anticancer effects. Because of that it can be confirmed as a plant of phytopharmaceutical importance.

KEYWORDS: *Passiflora foetida*, secondary metabolites, GC-MS, fruit pulp.

INTRODUCTION

Passiflora foetida (Linn.) is belonging to Passifloraceae family. It is native of tropical America and growing as wild in several parts of India. It is an herbaceous climber with the auxiliary tendrils and yellowish white hair on stems. *Passiflora* species are significant

because of their high therapeutic values. Tribes with traditional medicinal knowledge recommended *P. foetida* L. as a good source of high value pharmaceutical plant.^[1]

The whole plant is used in the treatment of insomnia and anxiety. The decoction of fruit is used for asthma and biliousness. Decoction of leaves and roots is emmenagogue and also used in hysteria.^[2] The major phytoconstituents in *P. foetida* were alkaloids, phenols, glycosides, flavonoids and cyanogenic compounds and passifloricins, polyketide and alpha-pyrone.^[3,4] *P. foetida* leaf extract is reported to possess anti-inflammatory effects and analgesic activity; anti-histamine; anti-depressant effects and estrogenic activity.^[5,6,7,8] The fruit's extract was reported to have anti-inflammatory effects and the whole plant possesses immunomodulatory effects.^[9,10] Other species of *Passiflora* is used as sedative, antidote for anxiety and hypertension.^[11] The aim of the present study was to find out the phytoconstituents present in seed and fruit pulp extracts by GC-MS analysis.

MATERIALS AND METHODS

Collection of Plant material and Preparation of Samples

The fruit of *P. foetida* L. was collected from Karur District, Tamilnadu, India. The fruit peel was removed completely and the seeds are separated from the pulps. The seeds were washed with sterile water, air dried. After complete drying, it was transferred into a mechanical grinder to make it fine powder. The seed and fruit pulp were used for cold extraction using methanol. The powder of seed and fruit pulp was soaked in methanol (1:6) for 24 hrs, separately. The extracts were then filtered through Whatman filter No.1. This procedure was repeated thrice. The filtrate were combined and then concentrated by using rotary evaporator at 40°C. The extract was stored in air tight container for further studies.

Qualitative analysis of Phytochemical

The methanolic extracts of seed and fruit pulp were used to evaluate the presence of alkaloids, flavonoid, terpenoids, tannin and saponin according to the methods described by Herborne.^[12]

Alkaloid

About 0.2 g of the extracts was warmed with 2% H₂SO₄ for two minutes, and then it was filtered and few drops of Dragendroffs reagent were added to the filtrate. Development of orange red precipitated indicated the presence of alkaloids.

Flavonoids

Extracts of about 0.2 g was dissolved in diluted NaOH and HCl was added drop wise. A yellow solution that turns colourless, indicated the presence of flavonoids.

Terpenoids

0.2 g of extracts was mixed with 2 ml chloroform (CHCl₃) and concentrated H₂SO₄ (3 ml) was added carefully. Formation of a layer of reddish brown color of the interface indicated positive result for the presence of terpenoids.

Tannins

Small quantity of extracts was mixed with water and heated on water bath. The mixture was filtered and ferric chloride was added to the filtrate. A dark green solution indicated the presence of tannins.

Saponin

About 0.2 g of the extract was shaken with 5 ml of distilled water and then heated to boil. Frothing (appearance of creamy of small bubbles) showed the presence of saponins.

GC MS Analysis

Methanolic extract of seed and fruit pulp were analyzed by using GC-MS (GC Clarius 500 Perkin Elmer) analyzer. The datas were obtained on an Elite-1(100% Dimethyl poly siloxane) column (30 0.25mm 1 μ mdf). Helium (99.999%) was used as the carrier gas with a flow rate of 1ml/min in the split mode (10:1).

Identification of Components

Compound which are present in the fruit pulp and seed extract was identified based on the molecular structure, molecular mass and calculated fragments. Interpretation on mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology (NIST). The spectrum of the unknown component was compared with the spectrum of the component stored in the NIST library version (2005), software, Turbomass 5.2.

RESULTS

The phytochemical tests on the crude methanolic extract of seed and fruit pulp of *P. foetida* exhibiting alkaloids, flavonoids, tannins, terpenoids, phenol and saponins were present and tabulated.

Table 1: Qualitative Phytochemical analysis of *Passiflora foetida* L. from Seed and Fruit Pulp Extract.

S. No.	Secondary Metabolites	Seed extract	Pulp extract
1.	Alkaloid	+	+
2.	Flavonoid	+	+
3.	Tannins	+	-
4.	Terpenoids	-	-
5.	Phenol	+	+
6.	Saponins	+	+

Key: + = present, - = absent

GC - MS analysis

GC MS analysis of seed and fruit pulp extracts of *P. foetida* L. revealed the presence of 12 and 15 phytoconstituents respectively (**Figure – 1 & 2**). The active compounds were identified based on their molecular weight (MW), retention time (RT) Molecular formula and concentration (peak area %) as presented in Table -2 and 3. In seed extract, major compounds like 11-Octadecenoic acid, methyl ester (31.82%), 9,12-Octadecadienoic acid, methyl ester (27.08%), Hexadecanoic acid, methyl ester (21.94%), Hexadecanoic acid, 15-methyl-, methyl ester (13.65%) and Phthalic acid , isobutyl octyl ester (2.85%) were prominent (Table – 2). Moreover, the most prevailing compounds of fruit pulp were Hexadecanoic acid, 15-methyl-, methyl ester (26.39%), 5-Hydroxymethylfurfural (25.18%), 2', 3'-Dideoxyribonolactone (11.89%), Levoglucosenone (11.21%), Furfura (6.76%); (Table – 3).

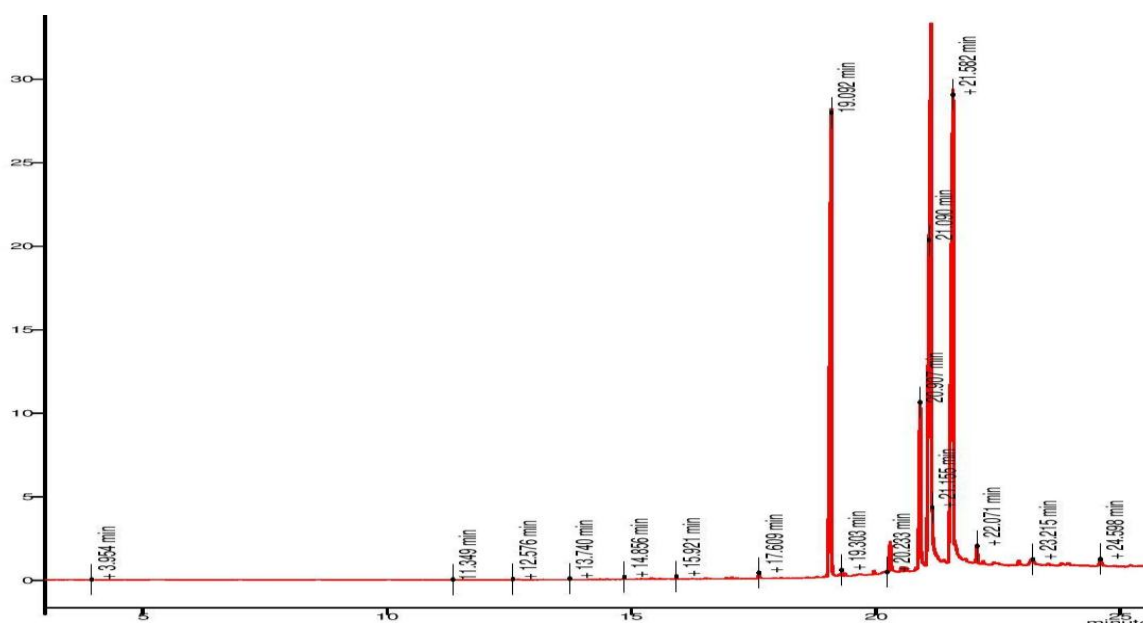


Fig. 1: GC-MS spectrum of seed extract of *P. foetida* L.

Table 2: Phyto - components identified in methanolic extract of seed of *P. foetida* by GC MS analysis.

S. No	R.T (min)	Compound Name	Molecular Formula	Molecular Weight	Peak Area (%)
1	11.34	Cyclopropanedodecanoic acid, 2-octyl-, methyl ester	C ₂₄ H ₄₆ O ₂	366	0.032
2	12.57	Octane, 2,4,6-trimethyl	C ₁₁ H ₂₄	156	0.143
3	14.85	Octane, 2,4,6-trimethyl	C ₁₁ H ₂₄	156	0.174
4	15.92	Oxalic acid, isobutyl nonyl ester	C ₁₅ H ₂₈ O ₄	272	0.681
5	16.95	Heptadecane, 2,6-dimethyl	C ₁₉ H ₄₀	268	0.470
6	17.67	Nonanoic acid, 9-oxo-, methyl ester	C ₁₀ H ₁₈ O ₃	186	0.840
7	19.09	Hexadecanoic acid, methyl ester	C ₁₇ H ₃₄ O ₂	270	21.94
8	19.33	11-Octadecenoic acid, methyl ester	C ₁₉ H ₃₆ O ₂	296	0.350
9	20.94	Hexadecanoic acid, 15-methyl-, methyl ester	C ₁₈ H ₃₆ O ₂	284	13.65
10	21.11	11-Octadecenoic acid, methyl ester	C ₁₉ H ₃₆ O ₂	296	31.82
11	21.58	9,12-Octadecadienoic acid, methyl ester	C ₁₉ H ₃₄ O ₂	294	27.08
12	22.07	Phthalic acid, isobutyl octyl ester	C ₂₀ H ₃₀ O ₄	334	2.850

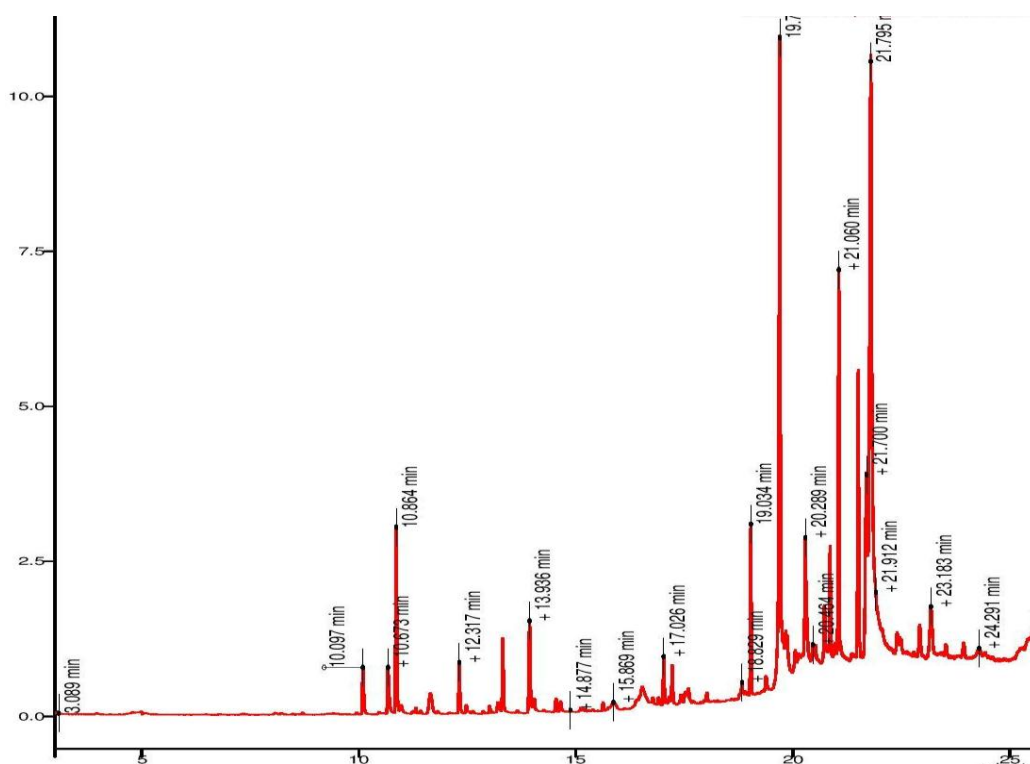


Fig. 2: GC-MS spectrum of fruit pulp extract of *P. foetida* L.

Table 3: Phyto –chemical components identified in the methanolic extract of fruit pulp of *P. foetida* L. by GC MS analysis.

S. No.	R. Time (min)	Compound Name	Chemical Formula	Molecular Weight	Peak Area (%)
1	10.09	Propargyl alcohol	C ₃ H ₄ O	56	01.31
2	10.86	Furfura	C ₅ H ₄ O ₂	96	06.76
3	12.31	2-Furancarboxaldehyde, 5-methyl	C ₆ H ₆ O ₂	110	01.31
4	13.25	1,3,5-Cycloheptatriene	C ₇ H ₈	092	03.14
5	13.93	Furan	C ₄ H ₄ O	68	01.48
6	17.02	Levogluosenone	C ₆ H ₆ O ₃	126	11.21
7	19.03	Hexadecanoic acid, 15-methyl-, methyl ester	C ₁₈ H ₃₆ O ₂	284	26.39
8	19.70	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl	C ₆ H ₈ O ₄	144	02.52
9	20.28	N-Aminopyrrolidine	C ₄ H ₁₀ N ₂	86	00.14
10	20.46	Hexadecanoic acid, 15-methyl-, methyl ester	C ₁₈ H ₃₆ O ₂	284	03.14
11	21.06	7-Hexadecenoic acid, methyl ester, (Z)	C ₁₇ H ₃₂ O ₂	268	01.25
12	21.50	11,14-Eicosadienoic acid, methyl ester	C ₂₁ H ₃₈ O ₂	322	00.88
13	21.70	2',3'-Dideoxyribonolactone	C ₅ H ₈ O ₃	116	11.89
14	21.79	5-Hydroxymethylfurfural	C ₆ H ₆ O ₃	126	25.18
15	23.18	11,14-Eicosadienoic acid, methyl ester	C ₂₁ H ₃₈ O ₂	322	03.40

In the present study, the phytochemicals were identified in methanol extract of *P. foetida* by GC-MS analysis and predict their biological activities (Table 4) based on Dr. Duke's Phytochemical and Ethnobotanical Databases created by Dr. Jim Duke of the Agricultural Research Service/USDA.

Table 4: Biological activity of identified chemical compounds from Fruit Pulp and seed Extracts of *P. foetida* L.

S. No.	Compound Name	Biological Activity
1.	Furfura	Antiseptic, Flavor, Fungicide, Insecticide, Irritant, Pesticide
2.	2-Furancarboxaldehyde, 5-methyl	Antimicrobial, Preservative
3.	Furan	Toxic
4.	Levogluosenone	Preservative
5.	Hexadecanoic acid, 15-methyl-, methyl ester	Antioxidant, Hypocholesterolemic, Nematicide, Pesticide, Antiandrogenic flavor, Hemolytic, Aldehyde reductase inhibitor
6.	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl	Antimicrobial, anti-inflammatory, anti-proliferative
7.	Hexadecanoic acid, 15-methyl-, methyl ester	Antibacterial and antifungal activity

8.	11,14-Eicosadienoic acid, methyl ester	Anti inflammatory, anti oxidant, anti arthritic, anti coronary.
9.	2',3'-Dideoxyribonolactone	Antimicrobial
10.	11-Octadecenoic acid, methyl ester	Anti inflammatory, Hypercholesterolemic, Nematicide, Cancer preventive, Hepatoprotective, Insectifuge, Antistaminic, Antiacne, 5 alpha reductase inhibitor, Antieczemic, Antiandrogenic, Anti coronary, Antiarthritic
11.	9,12-Octadecadienoic acid, methyl ester	Antiinflammatory, hypocholesterolemic, cancer preventive, hepatoprotective, nematicide, insectifuge, antihistaminic antieczemic, antiacne, 5-Alpha reductase inhibitor, antiandrogenic, antiarthritic, anticoronary, insectifuge
12.	5-Hydroxymethylfurfural	Antimicrobial Anti-inflammatory
13.	Nonanoic acid, 9-oxo-, methyl ester	Anti-microbial, anti-inflammatory, antitumor, anti hyperpigmentative, antiproliferative, anti-acne, cyto toxic, Anti-leukemic, oxy radical scavenging Activity.

DISCUSSION

The present study was carried out to evaluate the presence of phytoconstituents in seed and fruit pulp extract of *Passiflora foetida* by qualitative phytochemical analysis and GC-MS analysis. The study revealed that the presence of phytochemicals such as alkaloid, flavonoid, phenol, tannin and saponins, which are the major group of compounds responsible for antimicrobial, antioxidant and anticancer properties. The alkaloids, tannins and flavonoids are known to have curative activity against several pathogens such as *Escherichia coli*, *Salmonella typhi*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Streptococcus pyogenes* and *Bacillus subtilis*, therefore could be used for the treatment of various illnesses.^[13]

The present study was coincided Asir *et al.*, 2014^[14] reported that the seed extract of *P.foetida* possess 24 phytoconstituents. They also reported that 11-Octadecenoic acid, methyl ester, 9, 12-Octadecadienoic acid, methyl ester, Hexadecanoic acid, 15-methyl-, methyl ester and Phthalic acid, isobutyl octyl ester were present in the seed extract. Moreover, anti inflammatory compounds like Hexadecanoic acid, fragrance and flavoring agents such as 2-octenoic acid, 11, 14-Eicosadienoic acid, methyl ester etc are identified in the present investigation by GC-MS analysis. Hussain, *et al.*, 2008^[15], reported that hexadecanoic acid has anti inflammatory activity and 4H-Pyran-4-one, 2, 3-dihydro-3,5-dihydroxy-6-methyl has antimicrobial, anti-inflammatory and anti-proliferative activities.

Furthermore, the finding of the present study revealed that the fruit pulp and seed extracts of *P. foetida* contained chemical compounds such as Nonanoic acid, Hexadecanoic acid 9,12-Octadecadienoic acid, methyl ester 4H-Pyran-4-one and 2-Furancarboxaldehyde, which are responsible for antimicrobial activity, antioxidant and anticancer activity.

CONCLUSION

In the present work, the chemical constituents present in the methanolic extracts of seed and fruit pulp of *Passiflora foetida* L were evaluated. Based on the findings, was concluded that the presence of phytochemicals such as alkaloids, flavonoids, phenols, tannin and saponin were present in the extracts of seed and fruit pulp. Naturally occurring plant bioactive compounds are a great source, to treat various diseases. The extracts also possess biologically active constituents worthy, responsible for antibacterial, antioxidant and anticancer activity. Further study the purification of individual compounds in the methanolic extract of seed and pulp in needed to evaluate their bioactivities.

REFERENCES

1. Patil AS, Paikrao HM. Bioassay Guided Phytometabolites Extraction for Screening of Potent Antimicrobials in *Passiflora foetida* L., Journal of Applied Pharmaceutical Science, 2013; 2(9): 137-142.
2. Ebadi M. Pharmacodynamic Basis of Herbal medicine, Taylor and Francis group, Boca Raton, London, Newyork, 2007; 100.
3. Dhawan K, Dhawan S, Shaema A. *Passiflora* a review update. J Ethnopharmacol, 2004; 94: 1-23.
4. Echeverri F, Arango V, Quinones W, Terres F, Escobar G, Rosero Y. et al. Passiflorocins, Polyketides and pyrones from *Passiflora foetida* resin. Phytochemistry, 2001; 56: 881-885.
5. Sasikala V, Saravanan S, Parimelazhagan T. Analgesic and anti-inflammatory activities of *Passiflora foetida* L., Asian Pacific Journal of Tropical Medicine, 2011; 4(8): 600-603.
6. Chivapat S, Bunjob M, Shuaoprom A, Bansidhi J, Chavalittumrong P, Rangsrpipat A, Sincharoenpokai P. Chronic toxicity of *Passiflora foetida* L. extract. International Journal of Applied Research in Natural Product, 2011; 4(2): 24-31.
7. Santosh P, Venugopl R, Nilakash AS, Kunjbihari S, Mangala L. Antidepressant Activity of Methanolic Extract of *Passiflora foetida* Leaves In Mice, International Journal of Pharmacy and Pharmaceutical Sciences, 2011; 3(1): 112-115.

8. Michel BG, Koffib K, Stanislasa ZO, Alassanec T, Flaviena T. Oral acute toxicity and estrogenic effects of the extracts of *Passiflora foetida* Linn. (Passifloraceae) leaves in female Wistar albino rats, *Annals of Biological Research*, 2012; 3(9): 4609 - 4616.
9. Fernandez J, Noronha MA, Fernandes R. Evaluation of Anti-inflammatory activity of stems of *Passiflora foetida* Linn. in rats, *Research Journal of Pharmaceutical, Biological and Chemical Sciences*, 2013; 4(2): 1236-4.
10. Ranganatha N, Kuppast IJ, Veerashekar T, Kulkarni S. Assessment of Immunomodulatory Activity of Aerial Parts of *Passiflora foetida* L., *World Journal of Pharmacy and Pharmaceutical Sciences*, 2013; 2(3): 1176 - 1186.
11. Ingale AG, Hivrale AU. Pharmacological studies of *Passiflora* sp. and their bioactive compounds, *African Journal of Plant Science*, 2010; 4(10): 417-426.
12. Herborne JB. *Phytochemical Methods* 3rd Edn. Chapman and Hall Ltd., London, 1973; 135-203.
13. Hassan MM, Oyewale AO, Amupitan JO, Abdullahi, MS, Okonkwo EM. Preliminary Phytochemical and antibacterial investigation of crude extracts of the root bark of *Detarium microcarpum*. *J. Chem. Soc. Nigeria*, 2004; 29: 26-29.
14. Joseph Asir P, Priyanga S, Hemalakshmi S, Devaki K. In Vitro Free Radical Scavenging Activity and Secondary Metabolites in *Passiflora Foetida* L. *Asian Journal of Pharmaceutical Research and Health Care*, 2014; 6(2): 3-11.
15. Hussain AI, Anwar F, Sherazi STH, Przybylski R. Chemical composition, antioxidant and antimicrobial activities of basil (*Ocimum basilicum*) essential oils depends on seasonal variations, *Food Chemistry*, 2008; 108(3): 986–995.